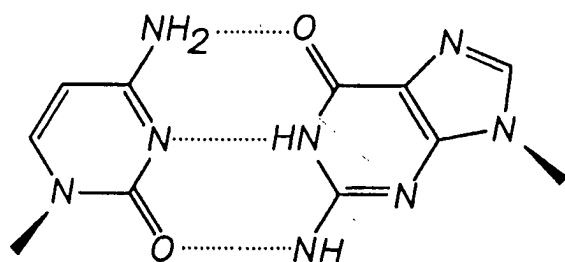
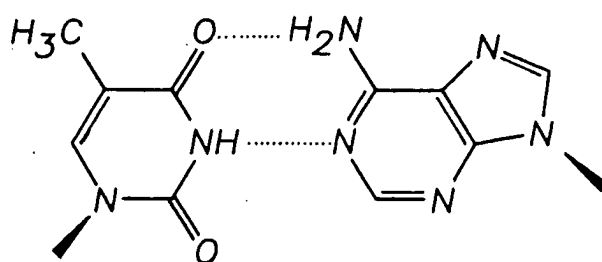


FIG. 1A



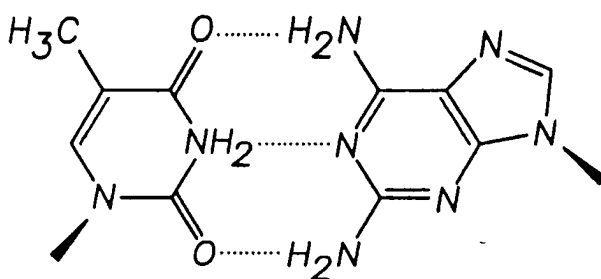
C•G

FIG. 1B



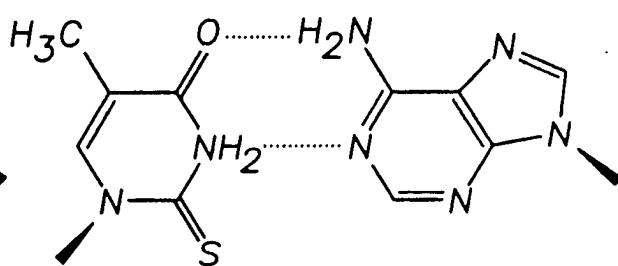
T•A

FIG. 1C



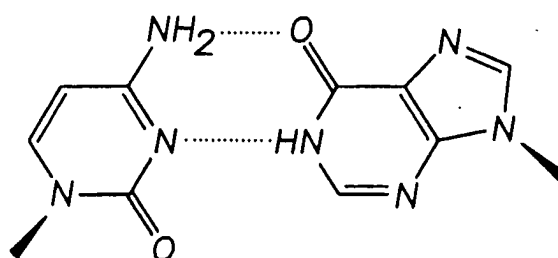
T•2-aminoA

FIG. 1D



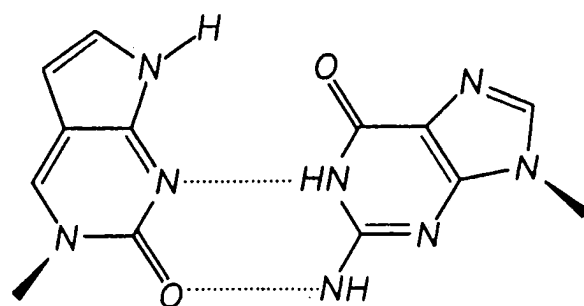
2-thioT•A

FIG. 1E



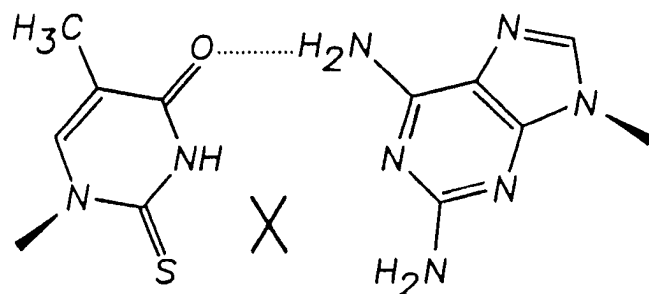
C•I

FIG. 1F



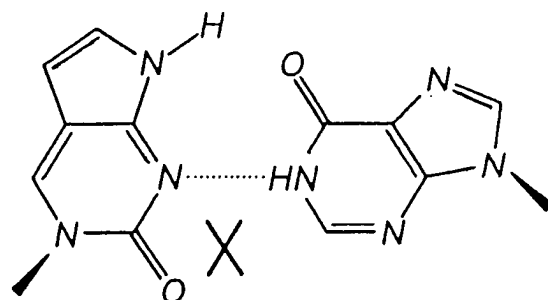
PyrroloPyr•G

FIG. 1G



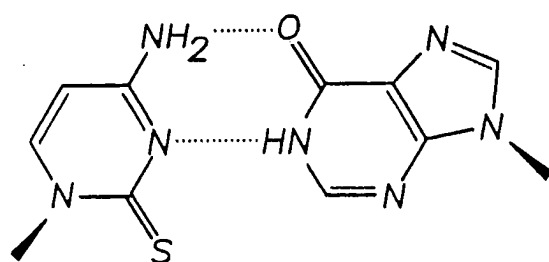
2-thioT•2-aminoA

FIG. 1H



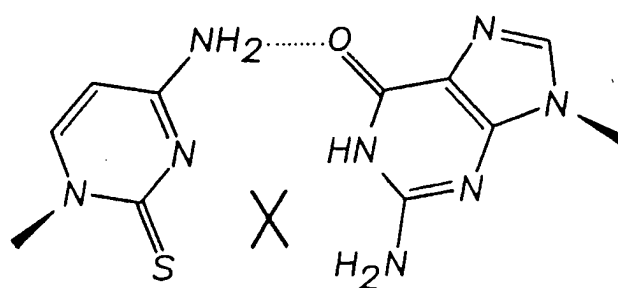
PyrroloPyr•I

FIG. 1I



2-thioC•I

FIG. 1J



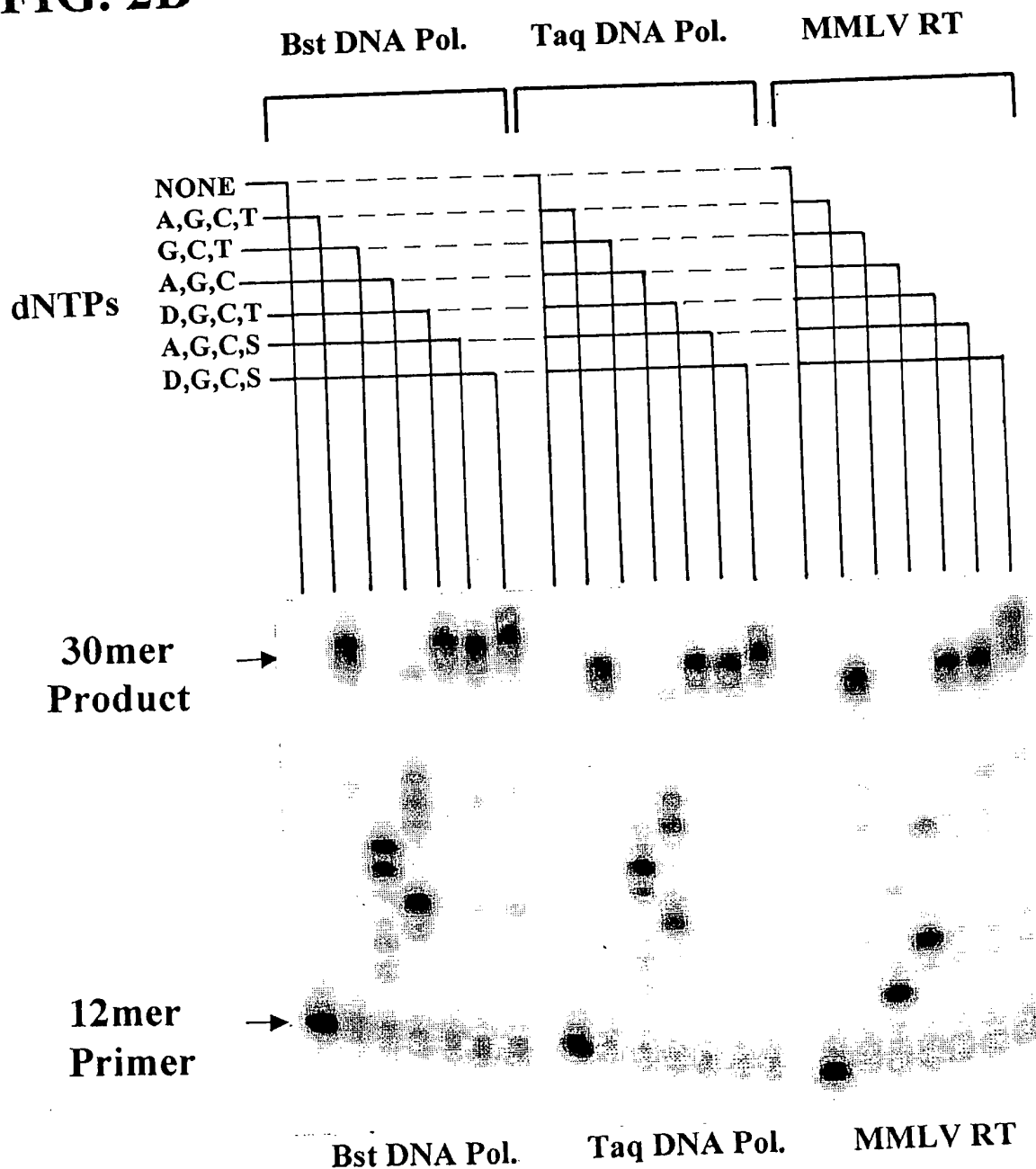
2-thioC•G

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FIG. 2A

5' -CGATAGGCTCTG →
3' -GCTATCCGAGACCCTGACTTGACACCTGTT-5'

FIG. 2B



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FIG. 3A

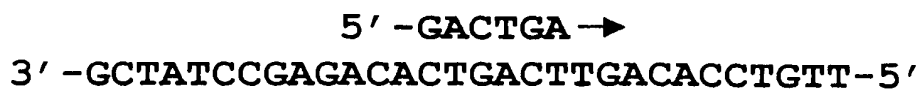
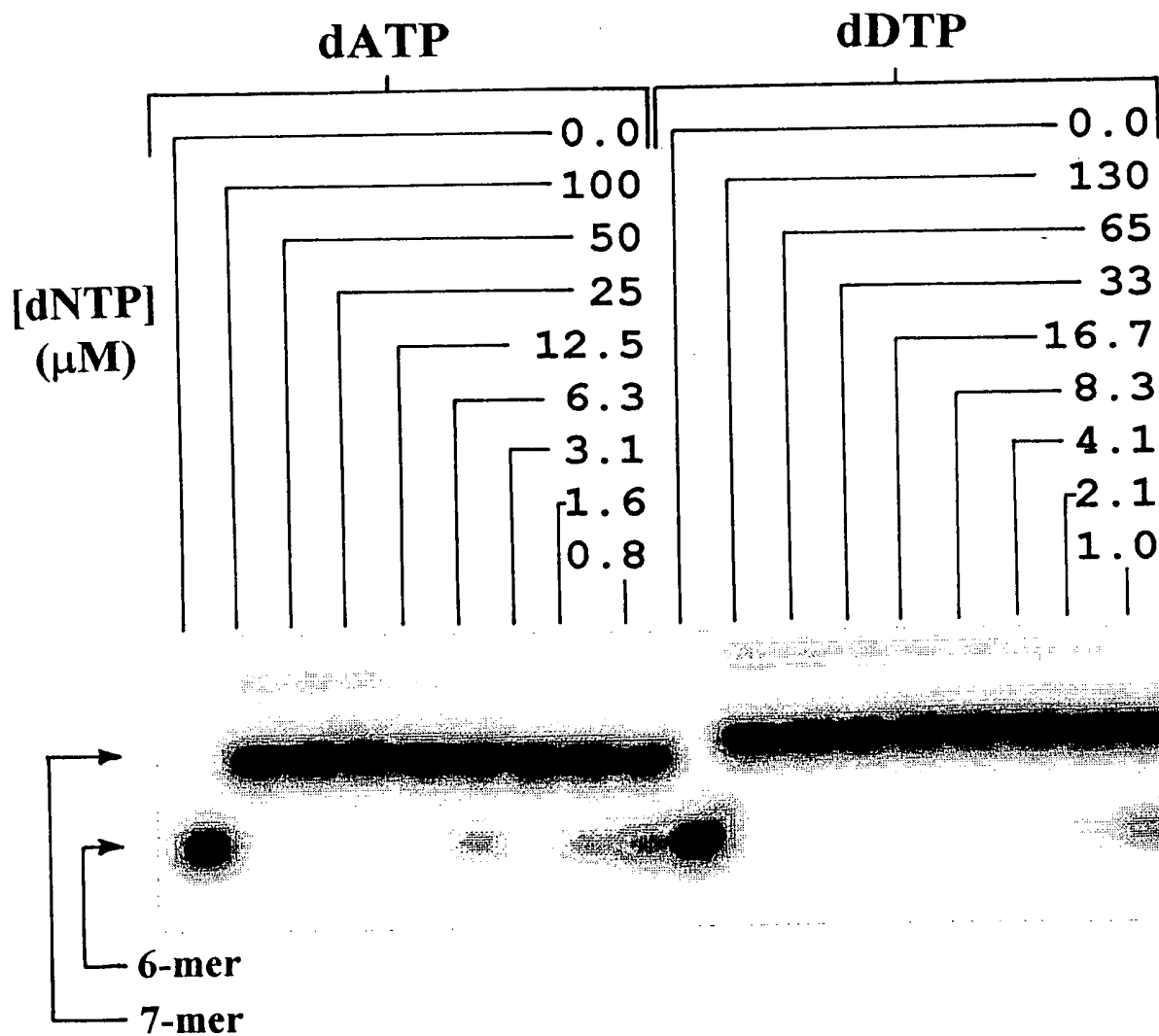
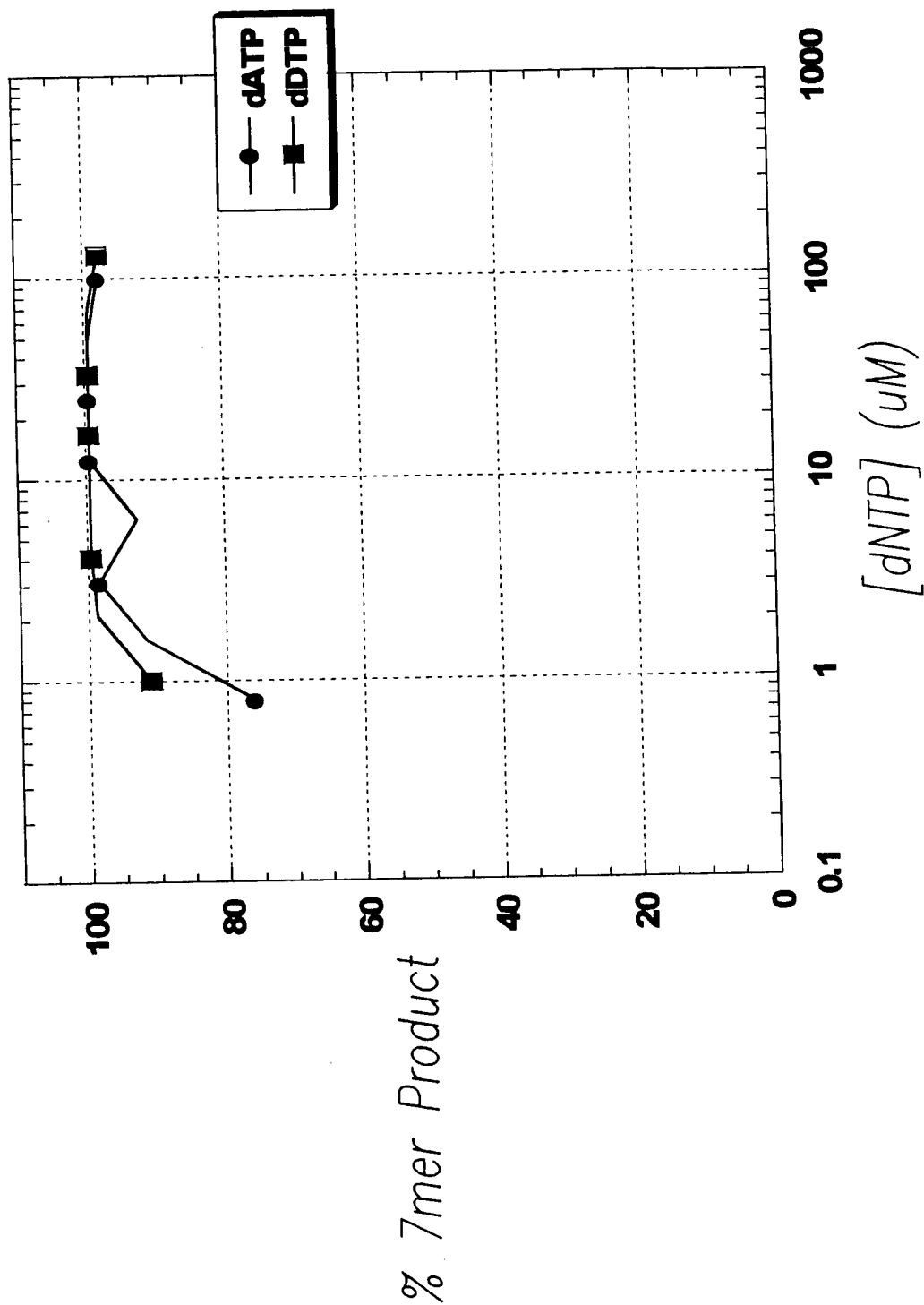


FIG. 3B



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FIG. 3C





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FIG. 4A

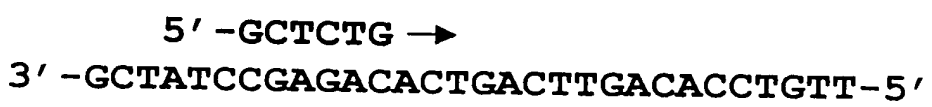
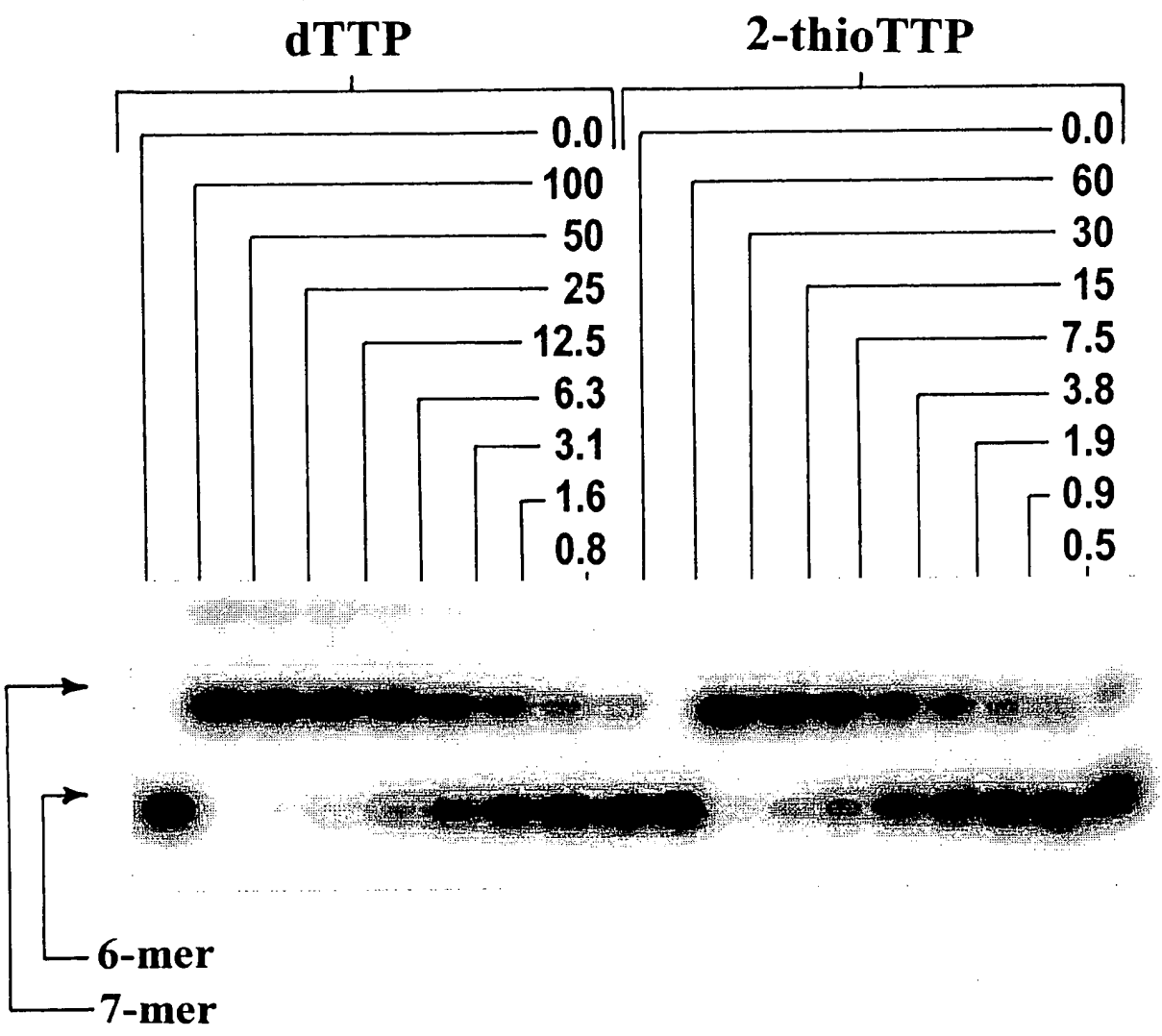


FIG. 4B



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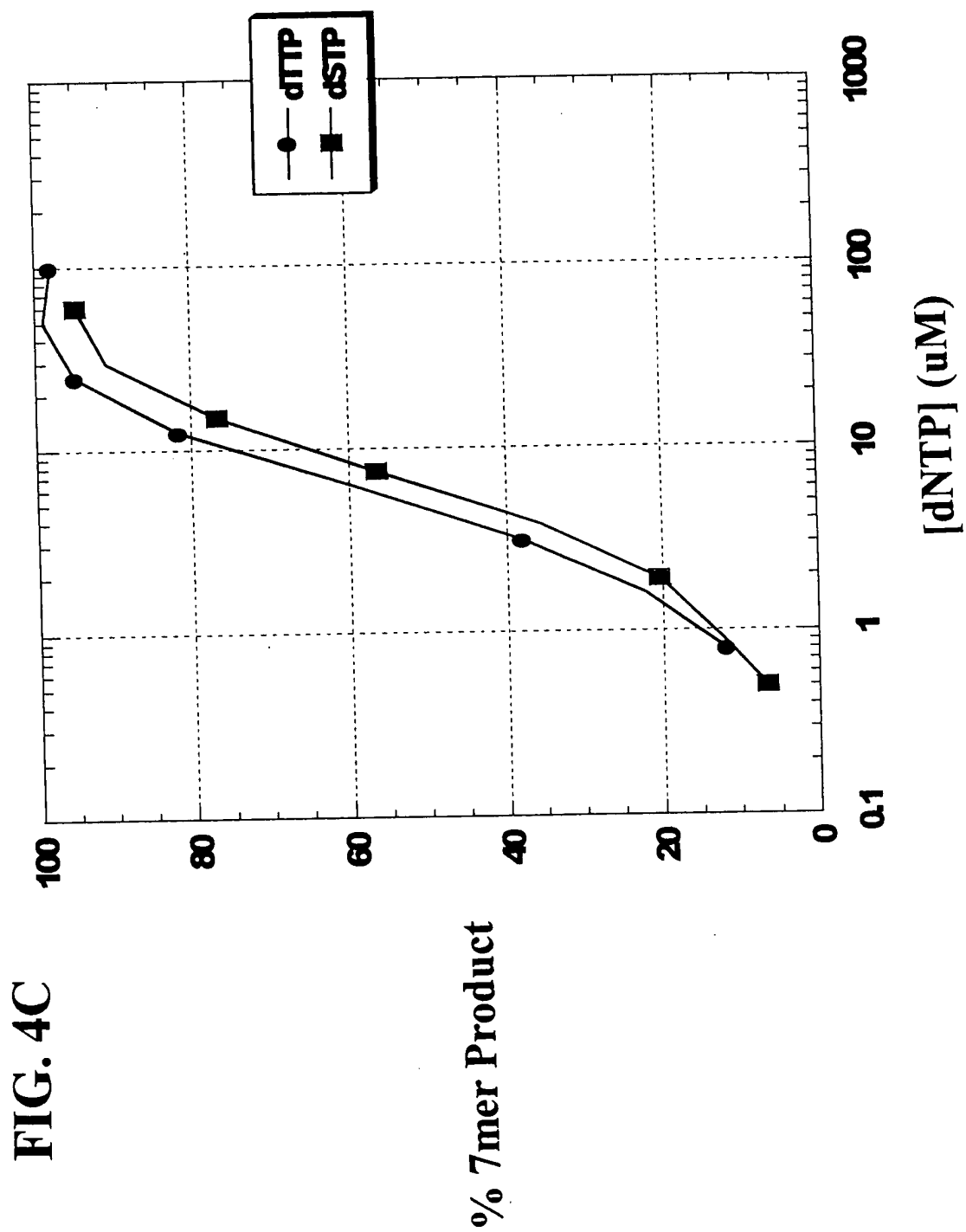


Fig. 5A

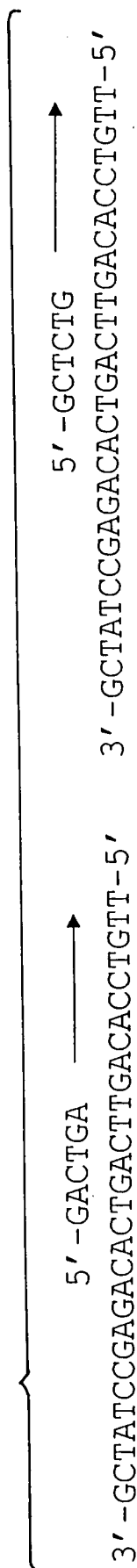


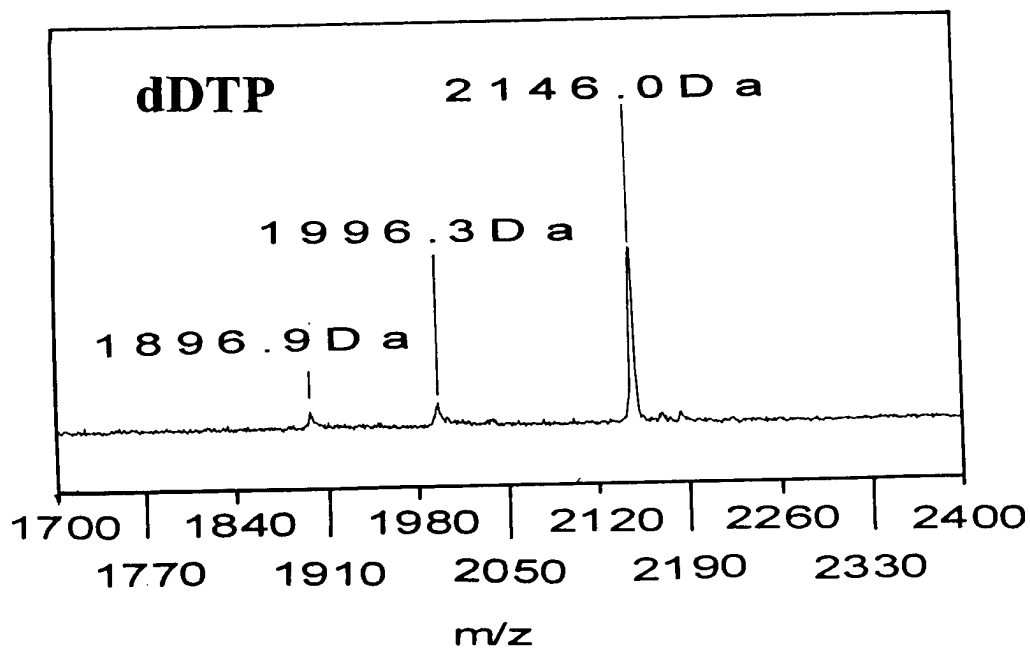
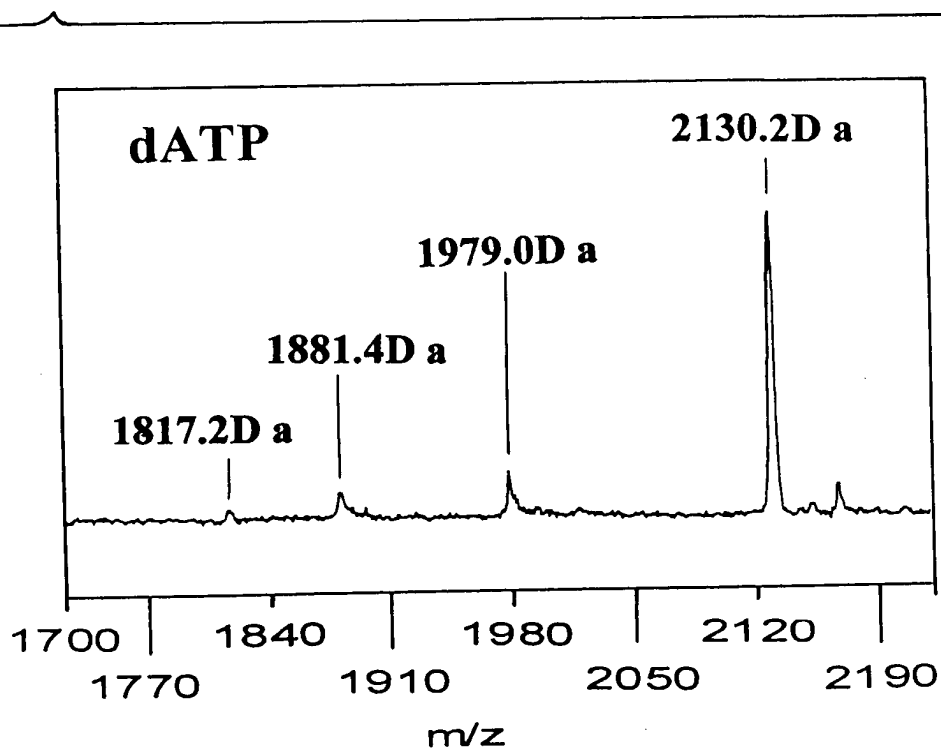
FIG. 5C

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Extension Nucleotide	X-mer	Predicted m/z (Positive Ion)*	Measured m/z (Positive Ion)	Predicted Δ m/z	Measured Δ m/z
None	GACTGA	1816.3	nd	--	--
dATP	GACTGAA	2129.5	2130.2	+15.0	+15.8
dTTP	GACTGAD	2144.5	2146.0		
None	GCTCTG	1783.2	1785.4 \pm 0.2	--	--
dTTP	GCTCTGT	2087.4	2089.7	+16.0	+16.4
d-2-thio-TTP	GCTCTGS	2103.4	2106.1		

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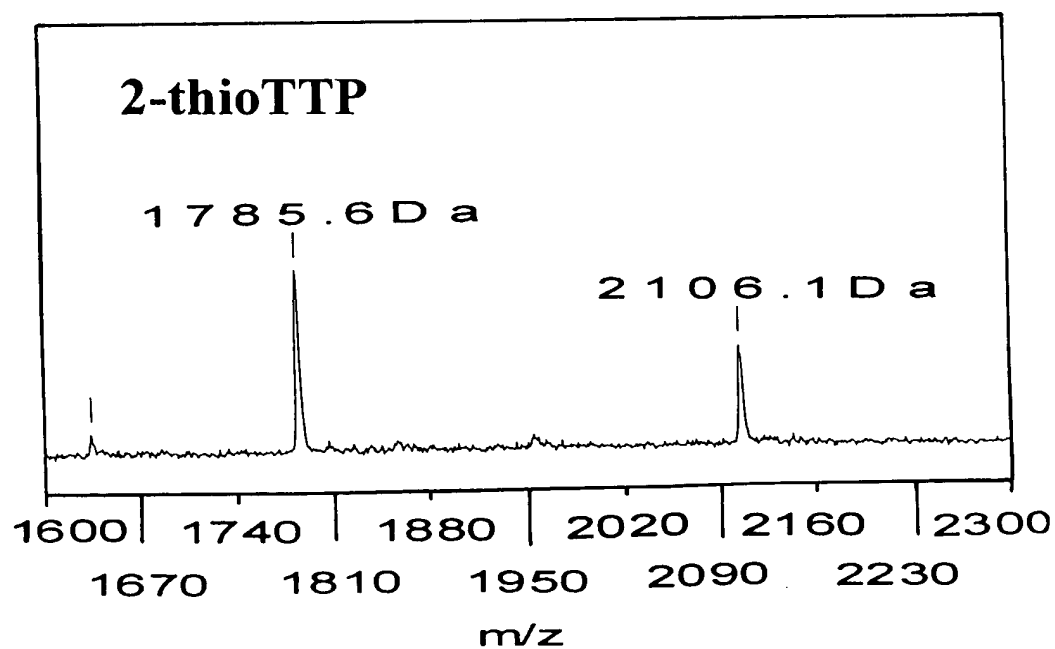
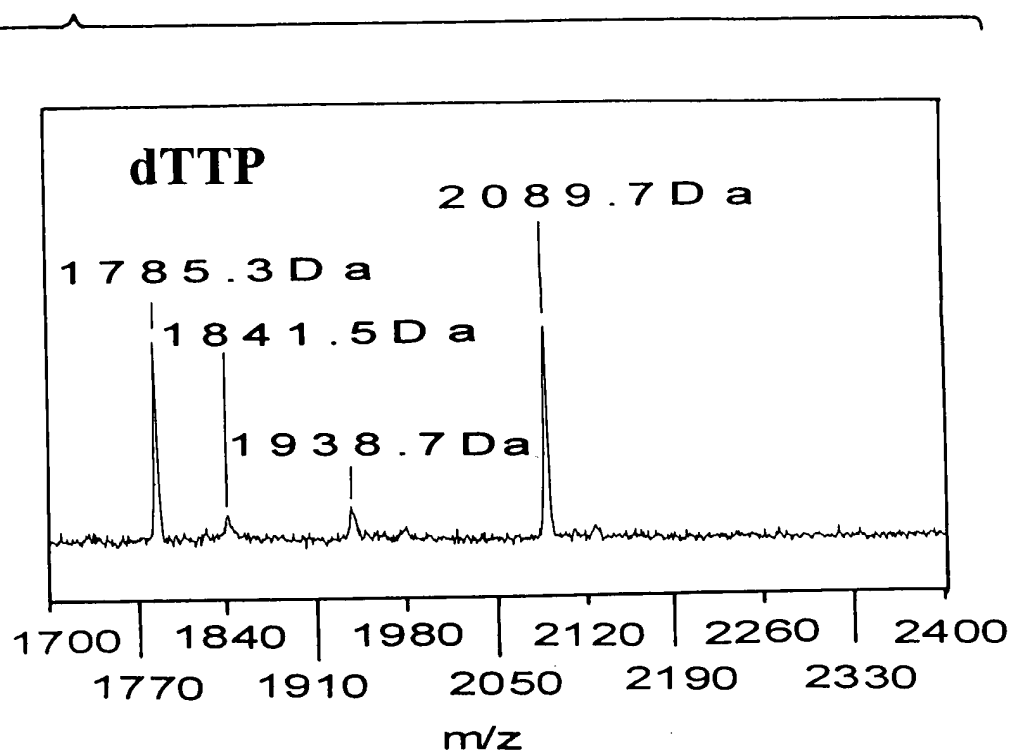
Fig. 5B-1





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Fig. 5B-2



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FIG. 6

5' - CTATCCGATCCATC →
3' - GATAGGCTAGGTAGTTCAAGTCAGAGCTTTGTCAGAGTTGTAAACAGGTGTTCGCATp-5'

dNTPs
Bst DNA Polymerase

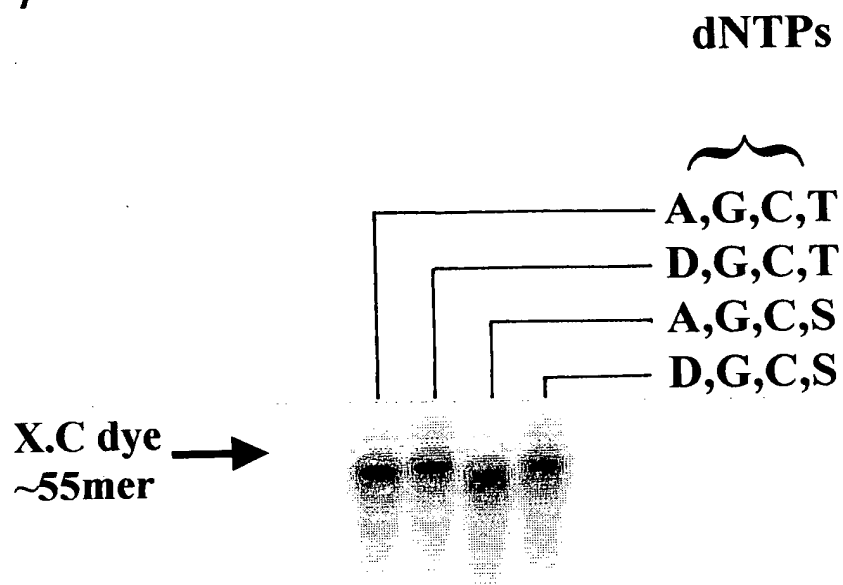
5' - CTATCCGATCCATCaagttcagttcgcgaaacagtcctcaacaatttgtccacagcgta
3' - GATAGGCTAGGTAGTTCAAGTCAGAGCTTTGTCAGAGTTGTAAACAGGTGTTCGCATp-5' →

λ Exonuclease

5' - CTATCCGATCCATCaagttcagttcgcgaaacagtcctcaacaatttgtccacagcgta-3'
+ pA, pG, pC pT

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FIG. 7



B.P dye →
~12mer

FIG. 8A

HP21AT

$$\begin{aligned}\Delta G^\circ &= -12.3 \text{ kcal/mole at } 37^\circ \text{C} \\ \Delta H^\circ &= -82.5 \text{ kcal/mole} \\ \Delta S^\circ &= -226.3 \text{ cal/} (^\circ \text{K} \cdot \text{mol}) \\ T_m &= 91.4^\circ \text{C}\end{aligned}$$

HP21DS

	10	20	
5' -CTATCCGATCCATCAA			G
		GT TCAGTCTC A	
		CAAGTCAGAG A	
3' -ATGCGACACCTGTTA			A
	50	40	30

10	20	
5'-CTATCCGATCCATCDD	G	
	GS CDGSC SC D	
	
	CDDGSCDGDG D	
		D
3'-DSGCGDCDCCGSSSD		30
		40
		50

FIG. 8B

HP26AT

$\Delta G^\circ = -3.8$ kcal/mole at 37 °C
 $\Delta H^\circ = -41.2$ kcal/mole
 $\Delta S^\circ = -120.5$ cal/(°K·mol)
 $T_m = 68.8^\circ\text{C}$

HP26DS

```

      10      20
5' -CTATCCGATCCATCAA      C   T   G
      GTT AG CTC A
      ||| ||| |||
      CAA TC GAG A
3' -ATGCGACACCTGTTTA      C   T   A
      50      40      30
  
```

```

      10      20
5' -CTATCCGATCCATCDD      C   S   G
      GSS DG CSC D
      |.. .| |..|
      CDD SC GDG D
3' -DSGCGDCDCCSGSSD      C   S   D
      50      40      30
  
```

FIG. 8C

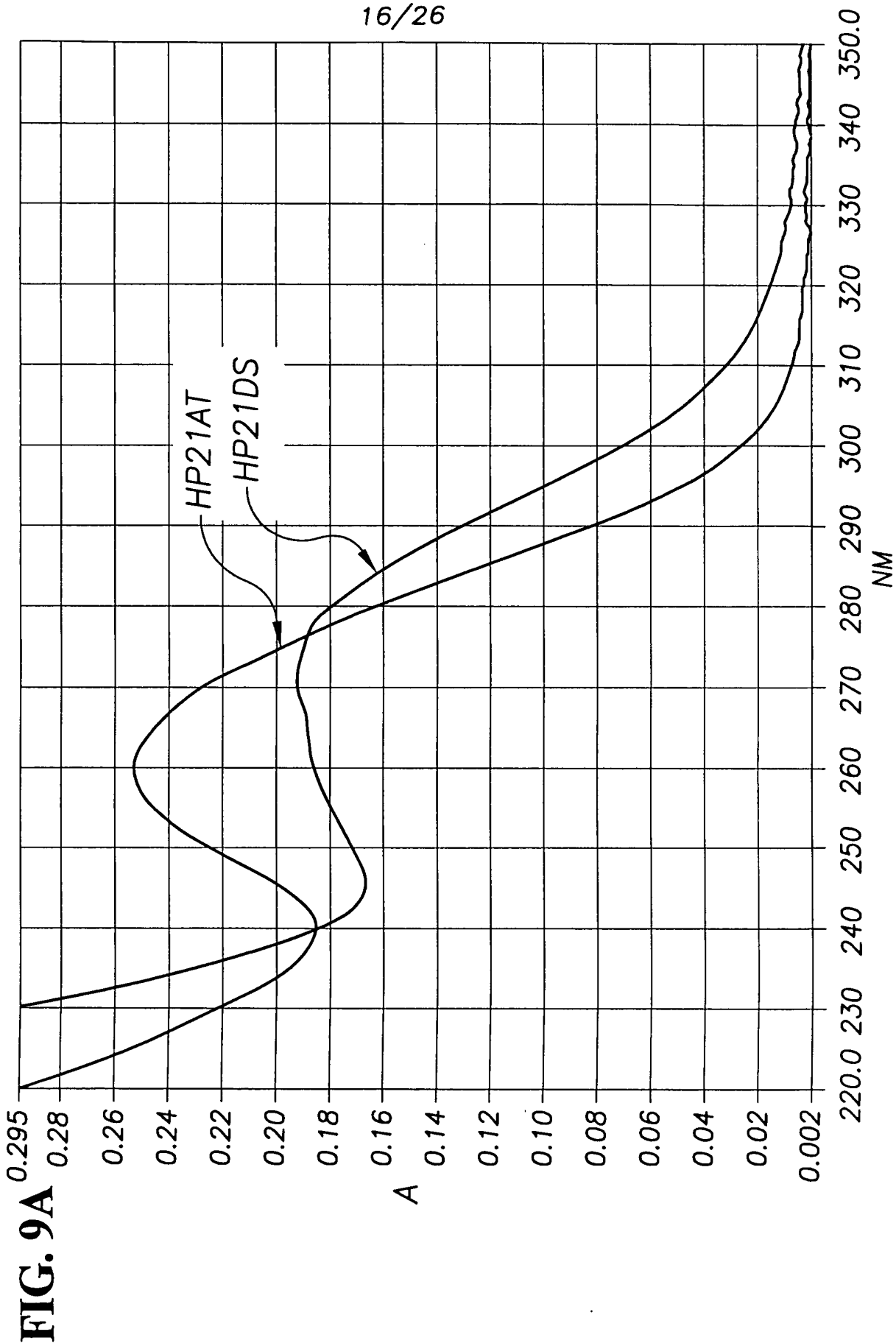
HP28AT

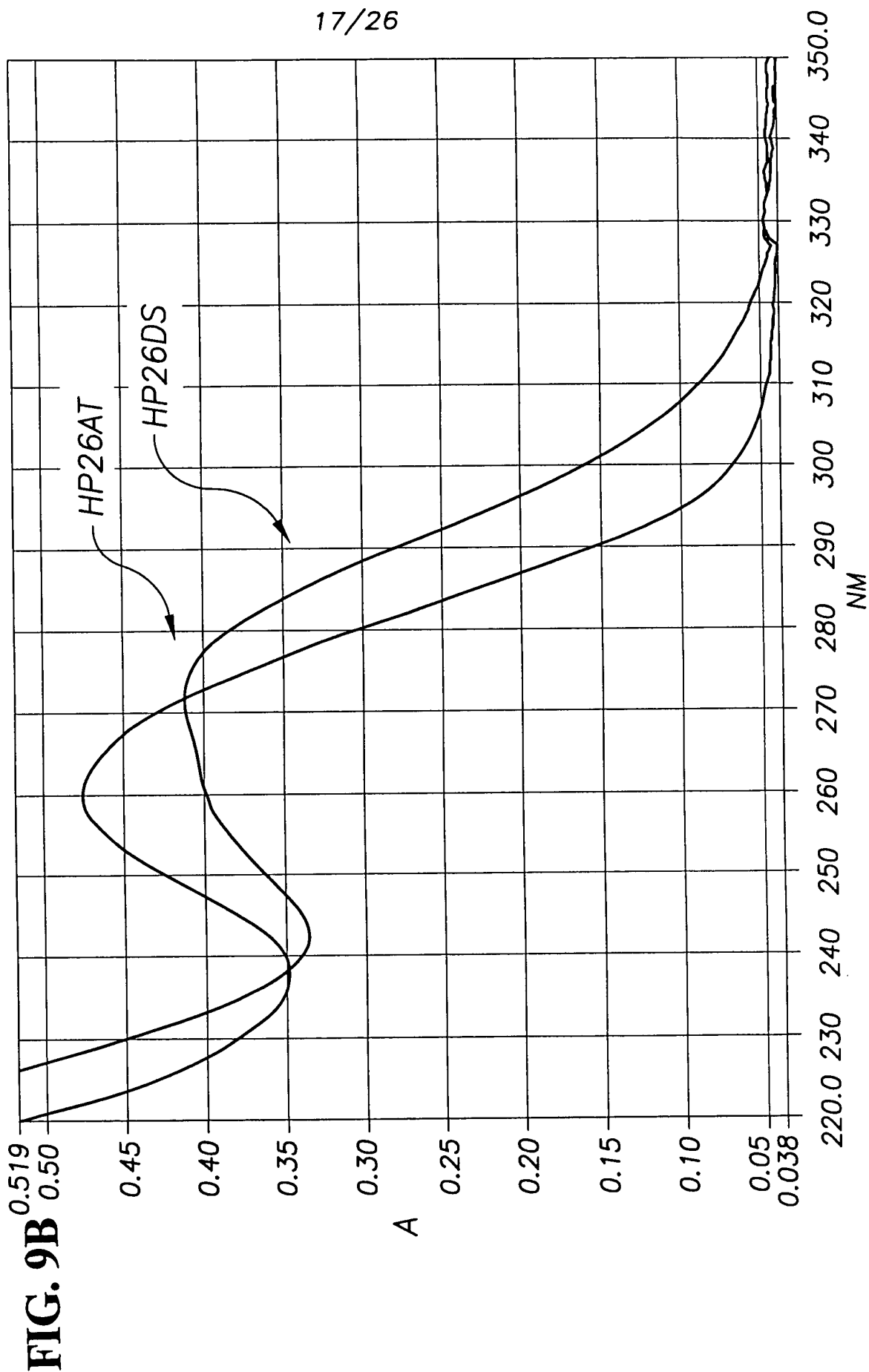
$\Delta G^\circ = 0.1$ kcal/mole at 37 °C
 $\Delta H^\circ = -27.4$ kcal/mole
 $\Delta S^\circ = -88.6$ cal/ (°K·mol)
 $T_m = 36.1^\circ\text{C}$

10	20	
5'-CTATCCGATCCATCAA	C T CG	
	GTT AG CT A	
	CAA TC GA A	
3'-ATGCGACACCTGTTA	C T CA	
50	40	30

HP28DS

10	20	
5'-CTATCCGATCCATCDD	C S CG	
	GSS DG CS D	
	
	CDD SC GD D	
3'-DSGCGDCDCCSGSSD	C S CD	
50	40	30





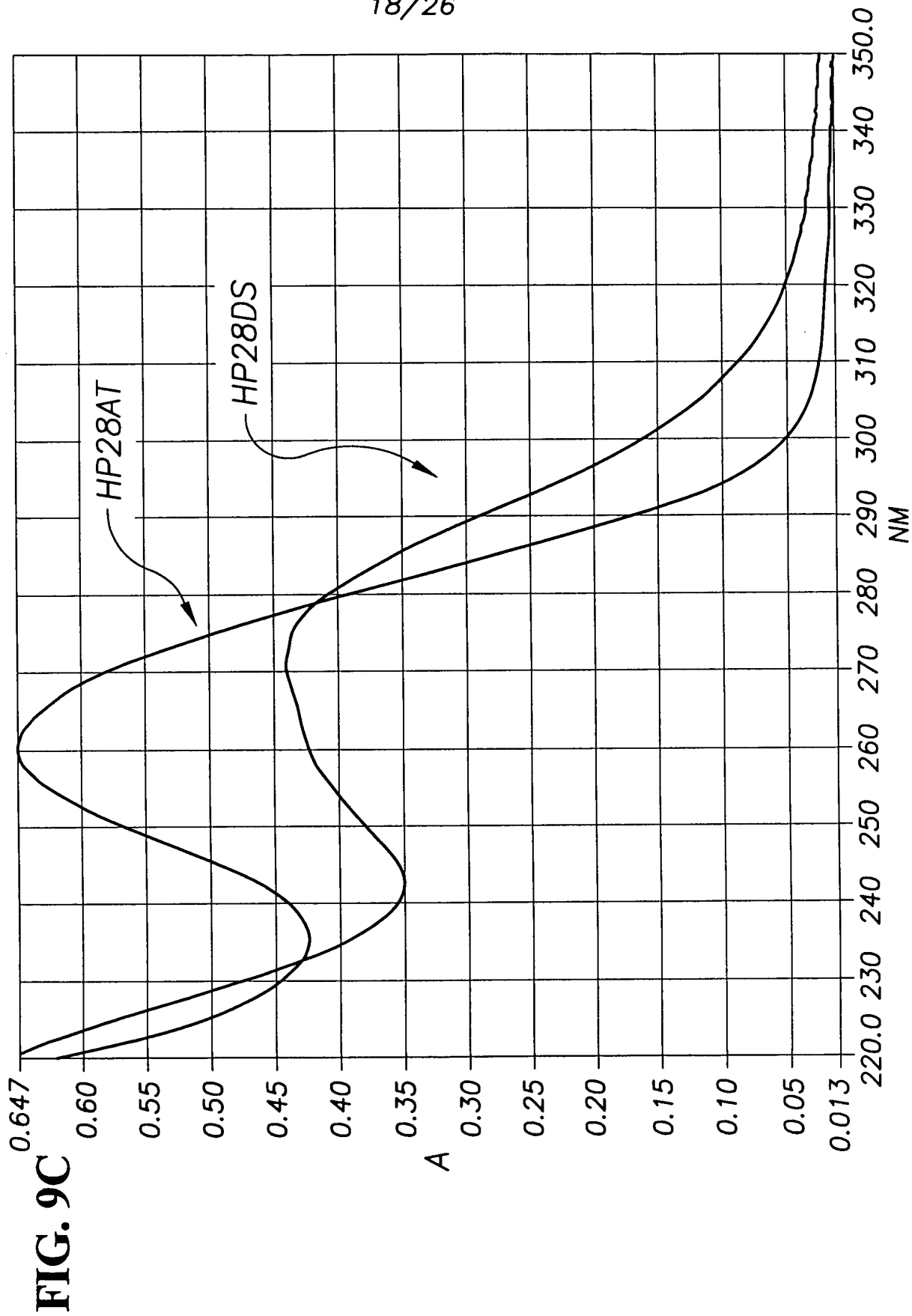
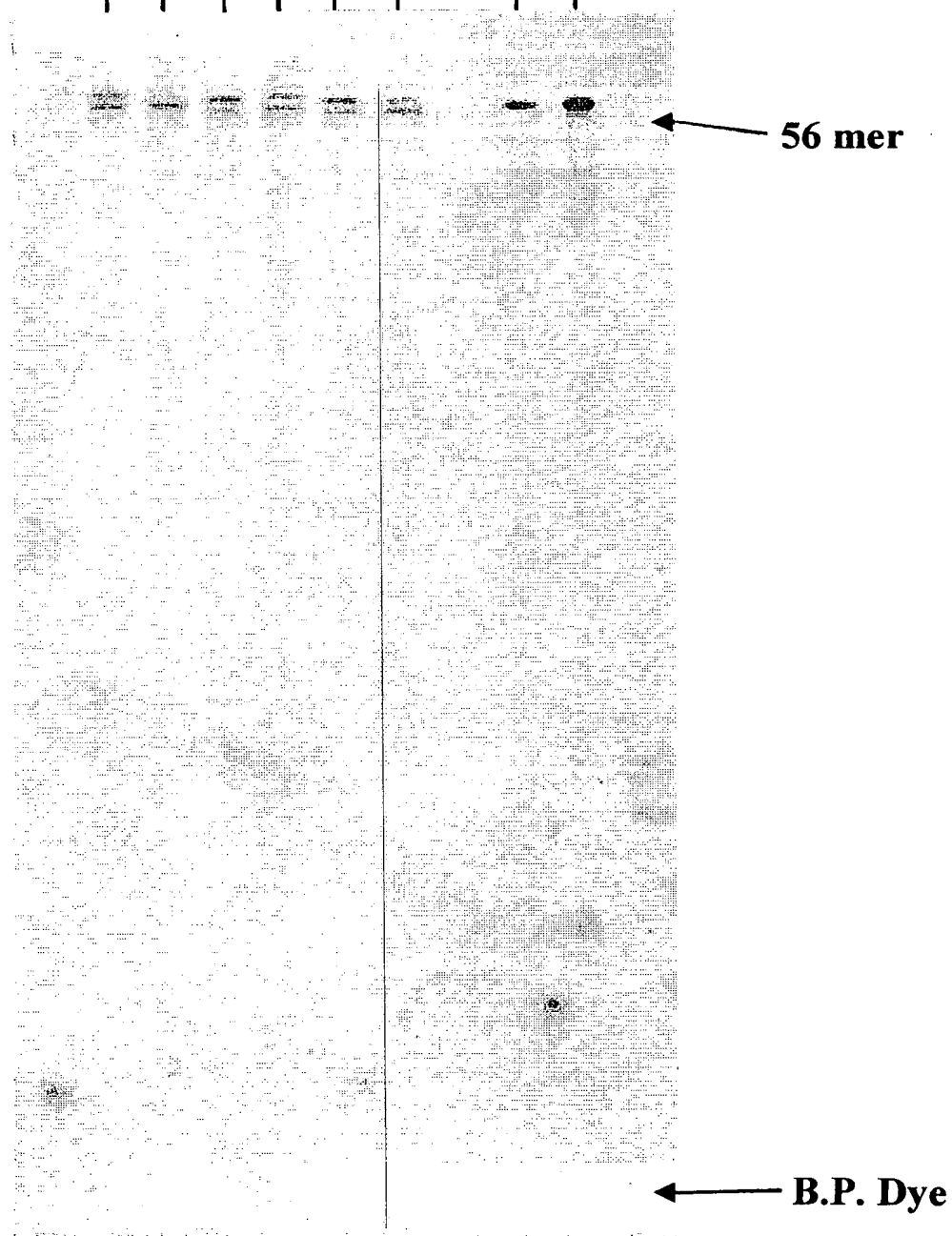


FIG. 10

HP21		HP26		HP28		HP21	
A	T	A	T	A	T	Template DNA	
DS	DS	DS	DS	DS	DS	DS	DS
0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2 (ug)



10% 7M Urea PAGE

FIG. 11A

← AGTCAGA 7mer-2169
 ← GTCAGA 6mer-543 ← CAGGTG 6mer-2978
 |||||

HP21 5'-CTATCCGATCCATCAAGTTCAGTCTCGAAAGAGACTGAACATTTGTCCACAGCGTA-3'

Target

← AGTCAGA 7mer-2169
 ← GTCAGA 6mer-543 ← CAGGTG 6mer-2978
 |||||

HP26 5'-CTATCCGATCCATCAAGTTCAGTCTCGAAAGAGTCTCAACATTTGTCCACAGCGTA-3'

Target

FIG. 11B

HP28 5'-CTATCCGATCCATCAAGTTCAGTCTCGAAACAGTCTCAACATTTGTCTCCACACGCGTA-3'
 Target

← AGTCAGA 7mer-2169
 ← GTCAGA 6mer-543
 |||||
 ← CAGGTG 6mer-2978
 |||||

TarZT 5'-TTGTCCACAGTTCAGTCTCAGAGCCTATCG-3'
 Target

← AGTCAGA 7mer-2169
 ← GTCAGA 6mer-543
 |||||
 ← CAGGTC 6mer-2978
 |||||

FIG. 12A

6-mer 543

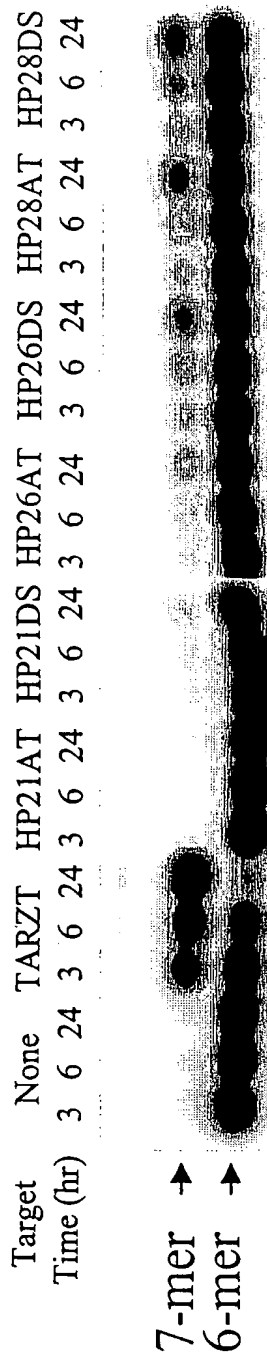
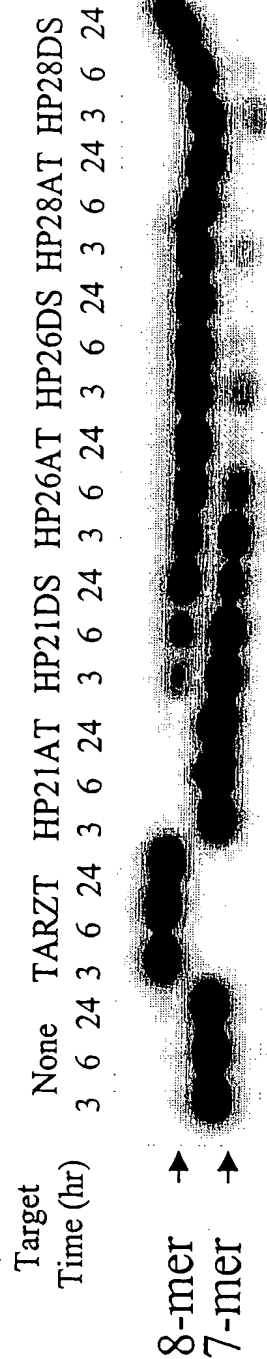


FIG. 12B

7-mer 2169



6-mer 2978

FIG. 12C

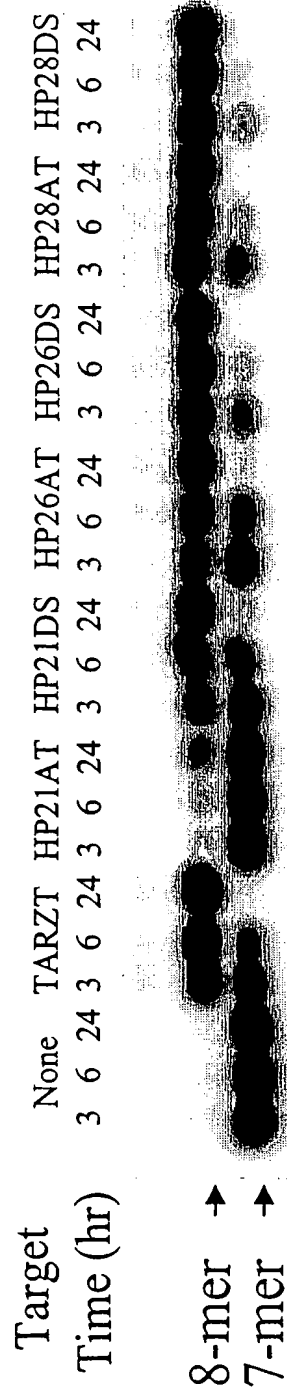
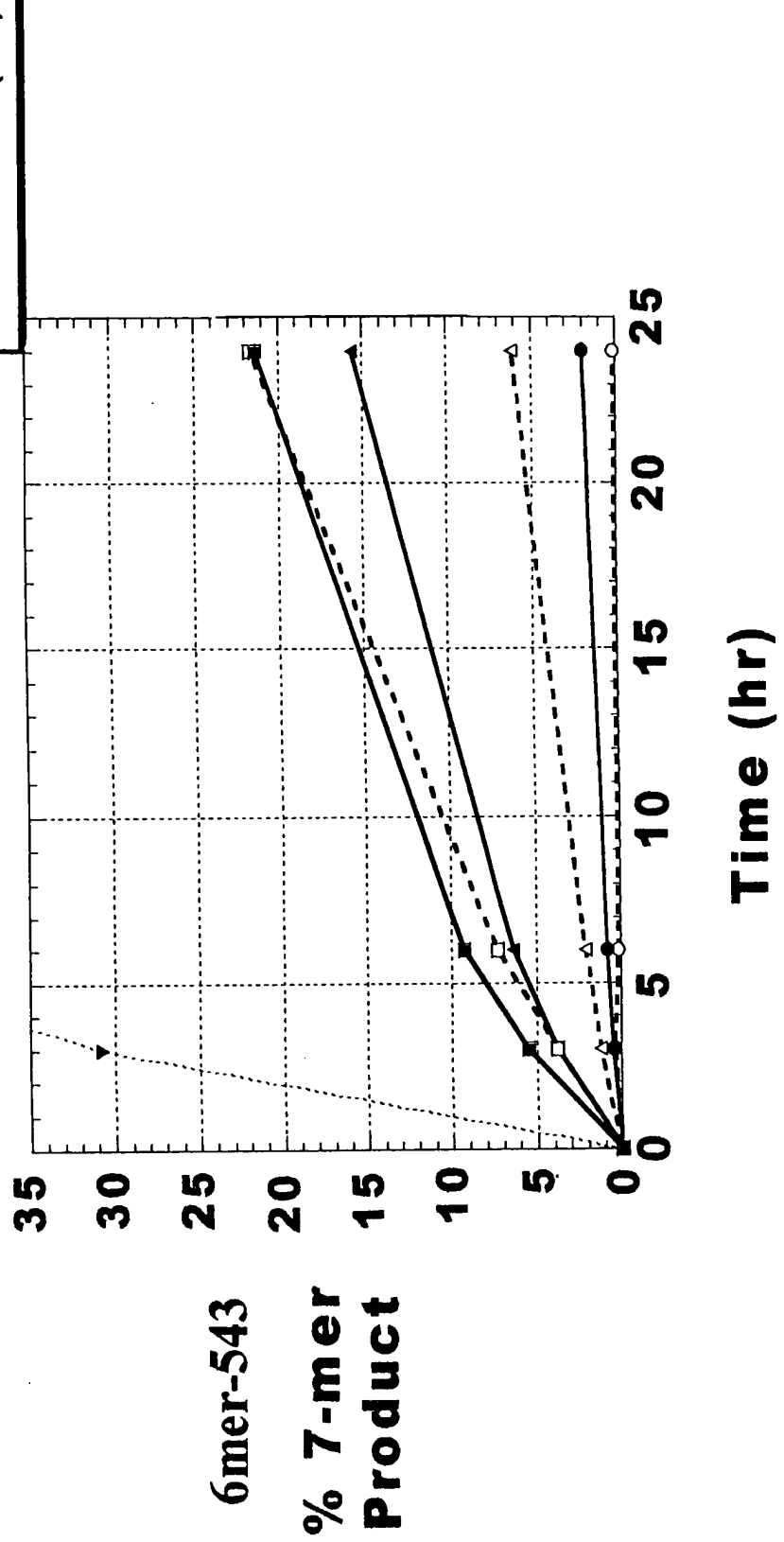


FIG. 13A



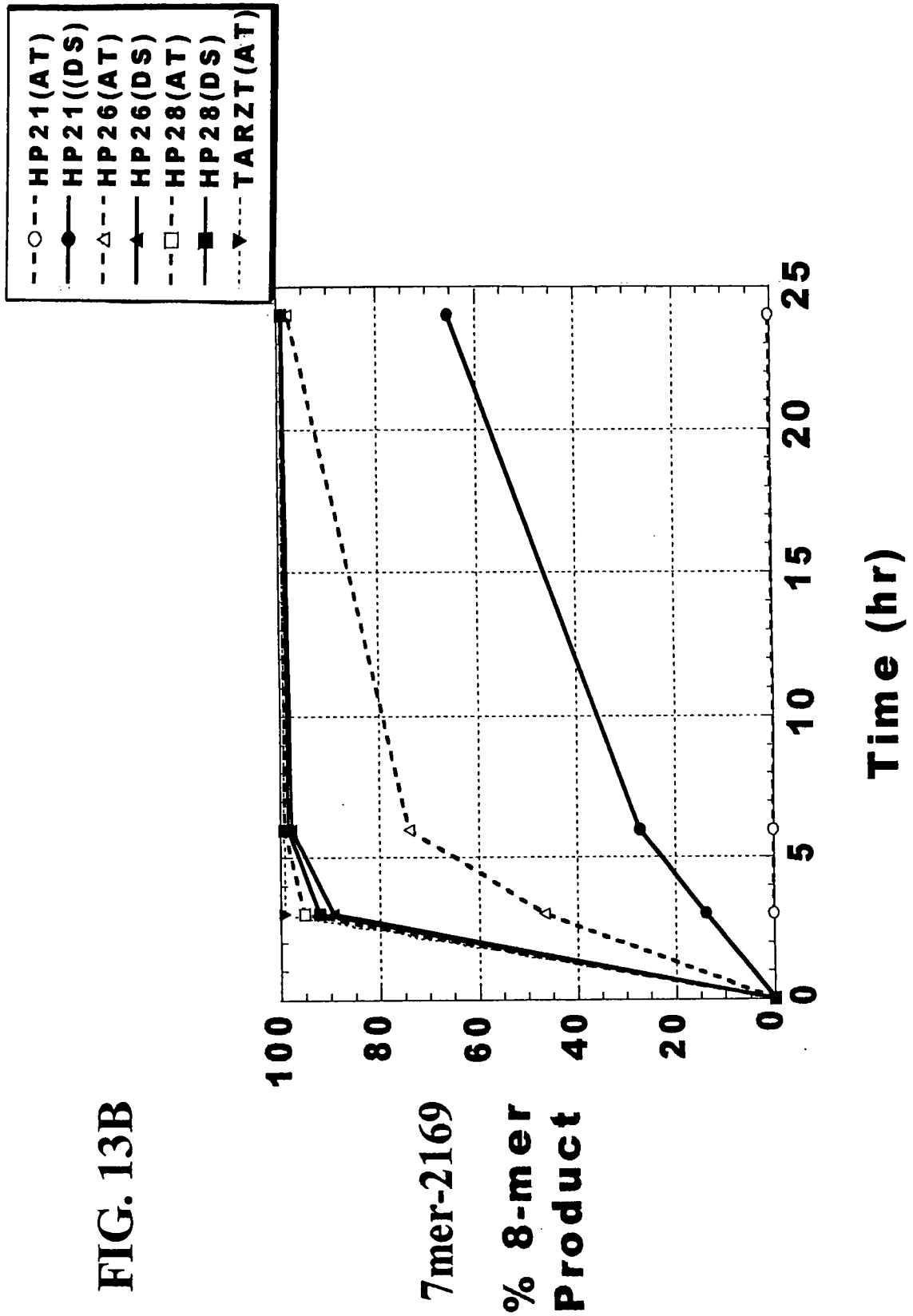


FIG. 13C

